CERTIFICATE OF FILING

I hereby certify that this paper and every paper referred to therein as being enclosed is being filed with the USPTO via facsimile to the designated fax number (571) 273-8300 or via EFS-Web and addressed to: Commissioner of Patents, P.O. Box 1450, Alexandria, VA 22313-1450,on the date below

Date: January 2, 2009

By /Kurt G. Briscoe/
Kurt G. Briscoe

Attorney Docket No. 106985-2
Confirmation No. 4429

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

APPLICANT	:	Gerold SCHULER et al
SERIAL NO.	:	10/618,134
CUSTOMER NO.	:	27384
FILED	:	July 11, 2003
FOR	:	$CD4^+CD25^-$ T CELLS AND TR1-LIKE REGULATORY T CELLS
ART UNIT	:	1644
EXAMINER	:	Amy Juedes

Hon. Commissioner of Patents
P.O. Box 1450
Alexandria, VA 22313-1450

DECLARATION UNDER 37 CFR §1.131

SIR:

We, the undersigned, hereby declare as follows:

1. We are the joint inventors of the subject matter disclosed and claimed in the above-identified application.

2. We completed the present invention in Germany prior to April 1, 2002.

3. To establish the date of completion of the invention claimed in this application, we submit true and correct reproductions of laboratory notebook entries for Experiment 336

("E336") as Exhibit A. Since these laboratory notebook entries are in the German language, we also submit a certified English language translation of these pages as Exhibit B.

4. From these laboratory notebook entries, it can be seen that the invention claimed in the above-identified application was made at least as early as March 2002.

5. The data from these laboratory notebook entries show that co-culture of $CD4^+CD25^-$ T cells with $CD4^+CD25^+$ T regulatory cells activated by stimulation with either anti-CD3/CD28 mAbs (graph on page 4) or mature allogenic DC (graph on page 6) anergized the $CD4^+CD25^-$ T cells, producing Tr-1-like regulatory cells that themselves exhibited suppressive properties.

6. The Tr-1 like regulatory cells exert their suppressive function via IL-10 as the addition of anti-IL-10 mAb abolished the suppressive function of these cells; see, again, the graphs on pages 4 and 6.

7. The suppressive function of $CD4^+CD25^+$ T regulatory cells, in contrast, was not abolished by the addition of anti-IL-10 mAb; see, again, the graphs on pages 4 and 6.

8. Fixation with formaldehyde also abolished the suppressive function of the Tr-1-like regulatory cells, whereas this was not the case for the $CD4^+CD25^+$ T regulatory cells; see, again, the graphs on pages 4 and 6.

9. The certified English language translation at various points indicates that certain words/entries could not be translated because they were illegible to the translator. These words/entries, although illegible to the translator, are legible to us from the original documents, and are translated as indicated in Exhibit C.

10. We further declare that all statements made herein of our own knowledge are true and that all statements made on information and belief are believed to be true; and that the foregoing statements were made with the knowledge that willful false statements and the like so made are punishable by fine or imprisonment, or both, under Section 1001 of Title 18 of the United States Code and that such willful false statements may jeopardize the validity of the application or any patent issuing thereon.

Dated: 18 Dec 2008 By _____
Gerold Schuler

Dated: 18 Dec 2008 By _____
Detlef Dieckmann

EXHIBIT A

TO RULE 131 DECLARATION OF THE INVENTORS

SCHULER ET AL. USSN 10/618,134

E 336 270079 1

20.03.02 * NAF von Len 270080 einen gr. Teil bekommen
gezählt: 2,5 · 10^8

* CD4 ∅ selektions - MACS

CD4+ $193 \cdot 10^6$ → CD25+ 3,2 Mio ; CD25⊝ 114 Mio

CD4∅ verworfen

~ 0,5 Mio/well - ideal.

* 48 well Pl. 1x-vivo 20 c. 5% S.
2x well 1 : CD4+ CD25 ∅ 0,8 Mio + CD4+ CD25 + 0,8 Mio
2x well 2 : CD4+ CD25+ 0,8 Mio
2x well 3 : CD4+ CD26- ∅ 0,8 Mio

je 1 well mit Anti CD3/28 10 µg/ml
je 1 well mit R-DC E284 1:20

37 °C O/N

* 96 well Pl. 1x-vivo 20 c. 5% S.
ca. 110 µl 1 well + IL2 10 U/ml

22.03.02 * 48 well Pl. v. 20.03.02 geerntet und gezählt:

	Ak CD3/28	R-DC
CD4+ CD25 +/- 1:1	1 Mio	1,1 Mio
CD4+ CD25 +	0,6 Mio	0,4 Mio
CD4+ CD25 ∅	0,4 Mio	0,75 Mio

CD4+ CD25 ∅ thawed : 102,6 Mio

* 1 PO E284 R-DC getaut: 1,6 Mio

* 98 U Pl 1x-vivo 020 c. 5% S. Rg.
CD4+ CD25 ∅ unbehandelt 1,6 Mio
CD4+ CD25 +/- 1:1 (CD3/28) 0,3 Mio
CD4+ CD25 +/- 1:1 (- " -) fix*1 0,15 Mio
CD4+ CD25 + (CD3/28) 0,3 Mio
– " – fix*1 0,15 Mio
CD4+ CD25 ∅ (CD3/28) 0,3 Mio

E336

(22.03.02) * 96 U Pl. / X-vivo 20C. 5% S.

	Reg.	
CD4+CD15∅ nehmd	1,6 Mio	+ RDC Exp. 284 1:20
CD4+CD15∅/+ 1:1 (R-DC)	0,3 Mio	
CD4+CD15∅/+ 1:1 (+-) fix	0,15 Mio	
CD4+ CD15+ (R-DC)	0,3 Mio	
CD4+ CD15∅ (R-DC)	0,3 Mio	
CD4+ CD15∅ (--) fix	0,15 Mio	

+ Anti J1210 10 µg/ml in einige wells (keine MLR - Photo well)

*[1] fixieren je 0,3 Mio - 0,5 Mio + 0,2 ml 2%. Formaldehyd ; 30 min 4°C waschen mit PBS

MLR-Protokoll

Exp No.: 336 T TC von: Leu TC pro well: 50000 DC von: / 3

		1	2	3	4	5	6	7	8	9	10	11	12
CD4+ CD15 Ø (ruhend)	A		Ø		CD4+ CD15 Ø + Il10 10µg/ml								
CD4+ CD15 Ø (ruhend)	B	CD4+ CD15+ / CD4+ CD15 Ø			CD4+ CD15 Ø fixiert		2 w wenig Zelle						
CD4+ CD15 Ø (ruhend)	C	CD4+ CD15+ / CD4+ CD15 Ø + Il10 10µg/ml											
CD4+ CD15 Ø (ruhend)	D	CD4+ CD15+ / CD4+ CD15 Ø fixiert											
CD4+ CD15 Ø (ruhend)	E	CD4+ CD15+											
CD4+ CD15 Ø (ruhend)	F	CD4+ CD25+ + Il10 10µg/ml											
CD4+ CD15 Ø (ruhend)	G	CD4+ CD15+ fixiert											
CD4+ CD15 Ø (ruhend)	H	CD4+ CD15 Ø											

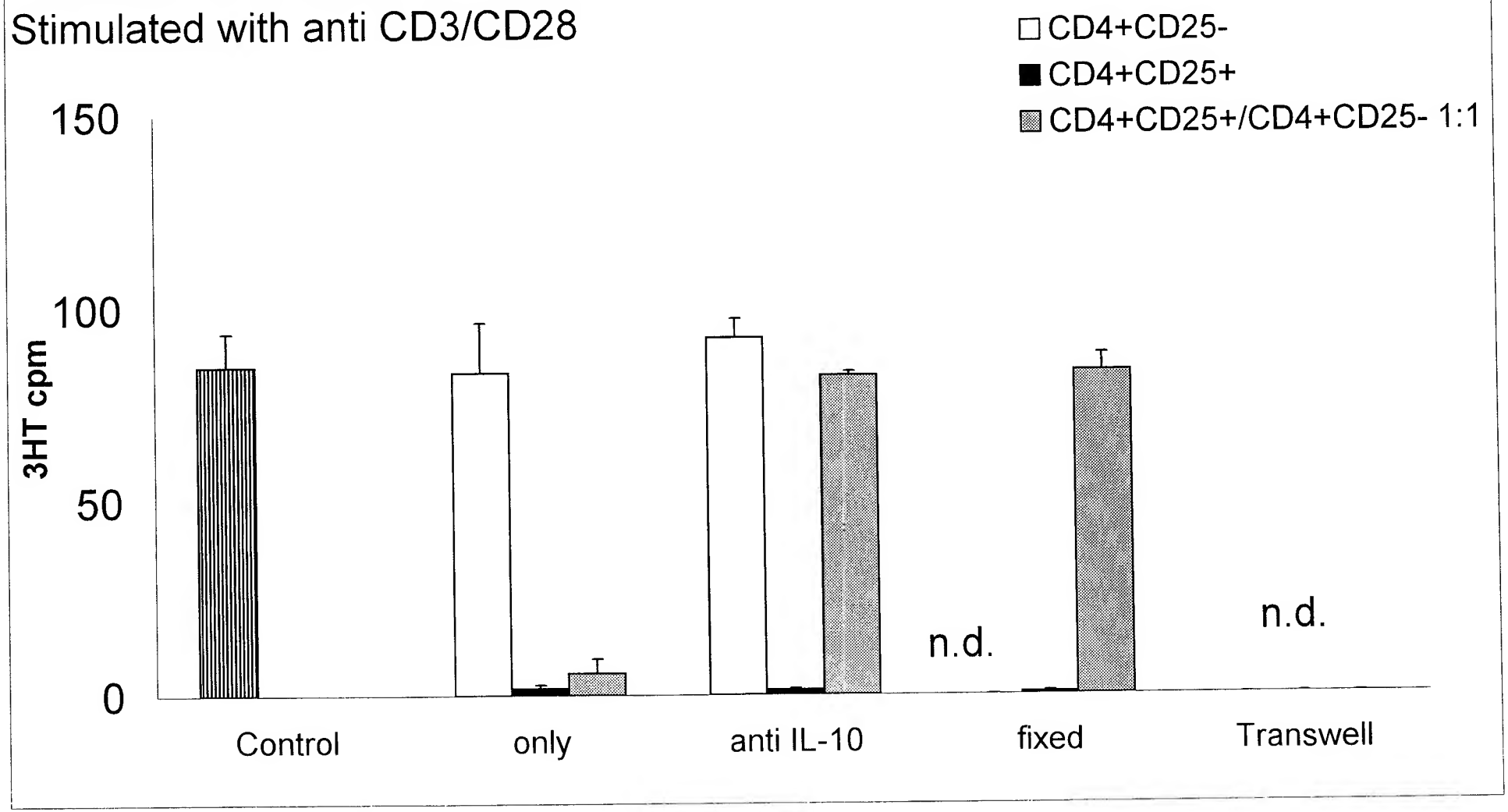
Angesetzt: _____ H^3- Zugabe: 26-03-02 Messung: 27.03.02 (3)

Exp. Beschreibung: + Anti CD3 10µg/ml 21.03.02

+ Anti CD28 -"- 22.03.02

Exp 336

Indicated Cells added to 10(5) CD4+CD25- T Cells
Stimulated with anti CD3/CD28



II

MLR-Protokoll

5

Exp No.: 336 TC von: Leu NHT TC pro well: 50 000 DC von: EZ84

		1	2	3	4	5	6	7	8	9	10	11	12
CD4+ CD15 ∅ (ruhend) R-DC 1:20	A		∅		CD4+ CD15 ∅ + J/10 10 μg/ml								
CD4+ CD15 ∅ (ruhend) R-DC 1:20	B	CD4+ CD15+ / CD4+ CD15 ∅ 1 : 1			CD4+ CD15 ∅ fixiert								
CD4+ CD15 ∅ (ruhend) R-DC 1:20	C	CD4+ CD15+ / CD4+ CD15 ∅ 1 : 1 + J/10 10 μg/ml											
CD4+ CD15 ∅ (ruhend) R-DC 1:20	D	CD4+ CD15+ / CD4+ CD15 ∅ 1 : 1 fixiert											
CD4+ CD15 ∅ (ruhend) R-DC 1:20	E	CD4+ CD15+											
CD4+ CD15 ∅ (ruhend) R-DC 1:20	F	CD4+ CD25+ + J/10 10 μg/ml											
CD4+ CD15 ∅ (ruhend) R-DC 1:20	G	~~CD4+ CD15+ fixiert~~		zu wenig Zellen									
CD4+ CD15 ∅ (ruhend) R-DC 1:20	H	CD4+ CD15 ∅											

Angesetzt: 22.03.02 H^3- Zugabe: 26.03.02 Messung: 27.03.02 (4)

Exp. Beschreibung:

Exp 336

Indicated Cells added to 10(5) CD4+CD25- T Cells

Stimulated with mature allogeneic DC

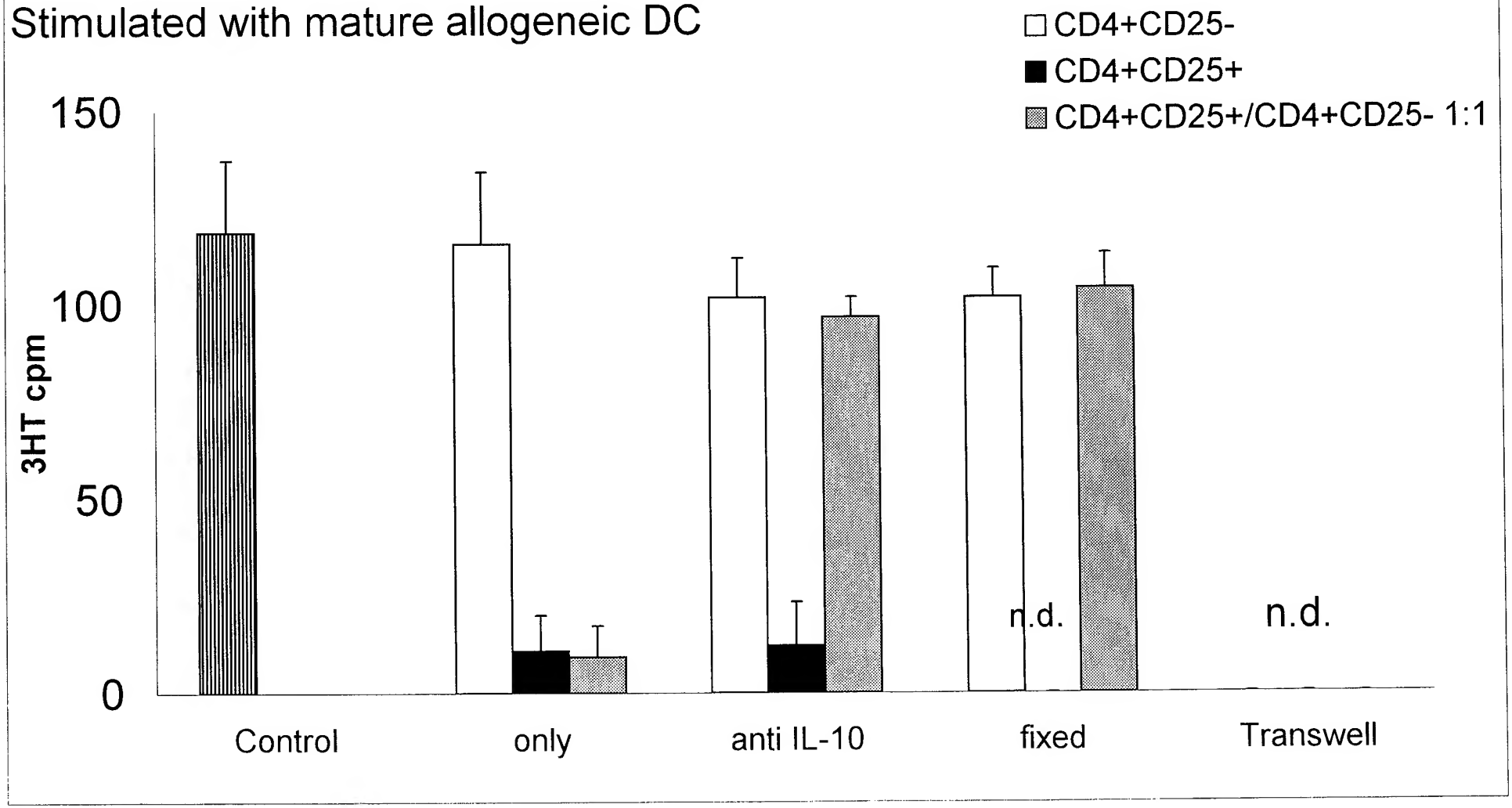


EXHIBIT B

TO RULE 131 DECLARATION OF THE INVENTORS

SCHULER ET AL.
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TRANSPERFECT

Certified Translation

ALBANY
AMSTERDAM
ATLANTA
AUSTIN
BARCELONA
BERLIN
BOSTON
BRUSSELS
CHARLOTTE
CHICAGO
DALLAS
DENVER
DUBAI
DUBLIN
FRANKFURT
GENEVA
HONG KONG
HOUSTON
IRVINE
LONDON
LOS ANGELES
MIAMI
MINNEAPOLIS
MONTREAL
MUNICH
NEW YORK
PARIS
PHILADELPHIA
PHOENIX
PORTLAND
RESEARCH TRIANGLE PARK
SAN DIEGO
SAN FRANCISCO
SAN JOSE
SEATTLE
SINGAPORE
STOCKHOLM
SYDNEY
TOKYO
TORONTO
VANCOUVER
WASHINGTON, DC

I, Angela Hsu, hereby declare that the following document is to the best of my knowledge and belief, a true and accurate translation of the document "E336," standing for, on information and belief, "Experiment 336," from German into English.

I further declare that all statements made herein of my own knowledge are true and that all statements made on information and belief are believed to be true; and further that these statements were made with the knowledge that willful false statements and the like so made are punished by fine or imprisonment, or both, under Section 1001 of Title 18 of the United States Code and that such willful false statements may jeopardize the validity of the application or any patent issued thereon.

By: Angela Hsu

Date: November 14, 2008

03/20/02 *NAF received a big part of Leu 270080
Counted: 2.5 * 10^8
*CD4∅ negative selection MACS

↓		↓
CD4+		CD4 ∅
193 · 10^6	Discarded	
↓		↓
CD25+		CD25-
3.2 million		114 million
~ 0.5 million/well	–	idem

* 48 well Pl. / x – vivo 20 c. 5% s.
2 x well 1 : CD4+ CD25∅ 0.8 million
+ CD4+ CD25+ 0.8 million
2 x well 2 : CD4+ CD25+ 0.8 million
2 x well 3 : CD4+ CD26- ∅ 0.8 million

1 well each with anti-CD3/28 10 µg/mL
1 well each with R-DC E284 1:20

37°C 0/N

* [illegible]6 well Pl. / x – vivo 20 c. 5% s
approx. 110 mL 1 well + [illegible] 10 u/mL

03/22/02 * 48 well Pl. of 03/20/02 tested and counted:

	Ab CD3/28	R-DC
CD4+ CD25+/- 1:1	1 million	1.1 million
CD4+ CD25+	0.6 million	0.4 million
CD4+ CD25 ∅	0.4 million	0.75 million

CD4+ CD25 ∅ resting: 102.4 million
* [illegible] E284 R-DC defrosted: 1.6 million
* [illegible] v Pl. / x – vivo 20 c. 5% s. reg.
CD4+ CD25 ∅ resting 1.6 million

CD4+ CD25+/- 1:1	(CD3/28)	0.3 million
CD4+ CD25+/- 1:1	(- "-) fix*[1]	0.15 million
CD4+ CD25+	(CD3/28)	0.3 million
- " -	fix *[1]	0.15 million
CD4+ CD25 ∅ (CD3/28) [illegible]		0.3 million

03/22/02	* 96 v Pl. / x – vivo 20 c. 5% s. reg.			
	CD4+ CD25 ∅ resting		1.6 million	
	CD4+ CD25 ∅/+ 1:1	(R-DC)	0.3 million	+ R-DC
	CD4+ CD25 ∅/+ 1:1	(- "-) fix	0.15 million	experiment
	CD4+ CD25+	(R/DC)	0.3 million	284
	CD4+ CD25 ∅	(R/DC)	0.3 million	1:20
	CD4+ CD25 ∅	- " - fix	0.15 million	
	+ anti IL-10 10 µg/mL in some wells (no MLR [illegible] well) *[1] fixation 0.3 million – 0.5 million each + 0.2 mL 2X formaldehyde; 30 min. at 4°C wash with PBS			

MLR protocol

I

Experiment number: *336* TC from: *Leu* TC per well: *50000* DC from: / *3*

		1 2 3 4 5	6 7 8 9	10 11 12
CD4+ CD25 Ø (resting)	A	*Ø*	*CD4+ CD25 Ø* *+ IL-10 10µg/mL*	
CD4+ CD25 Ø (resting)	B	*CD4+ CD25+ / CD4+ CD25 Ø* *1:1*	~~*CD4+ CD25 Ø*~~ ~~*too little [illegible]*~~ ~~*fixed*~~	
CD4+ CD25 Ø (resting)	C	*CD4+ CD25+ / CD4+ CD25 Ø* *1:1* *+ IL-10 1µg/mL*		
CD4+ CD25 Ø (resting)	D	*CD4+ CD25+ / CD4+ CD25 Ø* *1:1* *fixed*		
CD4+ CD25 Ø (resting)	E	*CD4+ CD25+*		
CD4+ CD25 Ø (resting)	F	*CD4+ CD25+* *+ IL-10 1µg/mL*		
CD4+ CD25 Ø (resting)	G	*CD4+ CD25+* *Too little cells* *fixed*		
CD4+ CD25 Ø (resting)	H	*CD4+ CD25 Ø*		

Prepared: Addition of H3: *03/26/02* measurement: *03/27/02* *3*

Description of experiment: *+ Anti CD3 10 µg/mL 03/21/02*
+ Anti CD18 -" - 03/21/02

[see source for graph]

MLR protocol

II

Experiment number: *336* TC from: *Leu [illegible]* TC per well: *50000* DC from: *E284* *5*

		1	2	3	4	5	6	7	8	9	10	11	12
CD4+ CD25 ∅ (resting) R-DC 1:20	A			*∅*				*CD4+ CD25 ∅ + IL-10 10µg/mL*					
CD4+ CD25 ∅ (resting) R-DC 1:20	B			*CD4+ CD25+ / CD4+ CD25 ∅ 1:1*				*CD4+ CD25 ∅ fixed*					
CD4+ CD25 ∅ (resting) R-DC 1:20	C			*CD4+ CD25+ / CD4+ CD25 ∅ 1:1 + IL-10 µg/mL*									
CD4+ CD25 ∅ (resting) R-DC 1:20	D			*CD4+ CD25+ / CD4+ CD25 ∅ 1:1 fixed*									
CD4+ CD25 ∅ (resting) R-DC 1:20	E			*CD4+ CD25+*									
CD4+ CD25 ∅ (resting) R-DC 1:20	F			*CD4+ CD25+ + IL-10 10 µg/mL*									
CD4+ CD25 ∅ (resting) R-DC 1:20	G			*CD4+ CD25+ too few cells fixed*									
CD4+ CD25 ∅ (resting) R-DC 1:20	H			*CD4+ CD25 ∅*									

Prepared: *03/22/02* Addition of H3: *03/26/02* measurement: *03/27/02* *4*

Description of experiment:

[see source for graph]

EXHIBIT C

TO RULE 131 DECLARATION OF THE INVENTORS

SCHULER ET AL.
USSN 10/618,134

Date	Illegible German Word/Entry	Translation
3/20/02 before "6 well"	9	9
3/20/02 before "10 µ/mL"	IL-2	IL-2
3/22/02 before "E284"	1 Zähv Gla	1 vial
3/22/02 before "v Pl."	96	96
3/22/02 after "(CD3/28)"	fix	fixed